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Impact of subchorionic hematoma on maternal serum alpha-fetoprotein levels and second trimester screening outcomes in threatened abortion cases

Tugrul Basar[1], Abdurrahman Alp Tokalioglu[1], Rıza Dur[1], Tufan Arslanca[2], Metin Altay[1]
[1] Department of Obstetrics and Gynecology, Faculty of Medicine, University of Health Sciences, Etlik Zubeyde Hanim Women's Health Training and Research Hospital
[2] Department of Obstetrics and Gynecology, University of Health Sciences, Ankara Bilkent City Hospital, Ankara, Turkey

Abstract

Aim: This study aims to examine the effects of subchorionic hematoma (SCH) on maternal serum alpha-fetoprotein (MSAFP) levels and the outcomes of second-trimester triple screening tests in patients who were diagnosed with threatened abortion during the first trimester.

Material and Methods: A retrospective study involved 922 patients who were diagnosed with threatened abortion between August 2013 and August 2015. After excluding cases of abortion, lack of follow-up, and those that did not meet inclusion criteria, 435 patients were included. Out of these, 102 had SCH (Group 1) while 248 did not (Group 2). Data collected comprised demographic characteristics, ultrasound findings, and triple screening test results. Statistical analyses included the use of student's t-test, chi-square test, and correlation analysis. Additionally, ROC analysis was conducted to evaluate the diagnostic value of SCH as a predictor for elevated MSAFP levels.

Results: The patients had an average age of 27.5 ± 5.6 years, and the mean gestational age at the time of diagnosis was 10.8 ± 3.1 weeks. Patients with SCH showed significantly higher MSAFP levels compared to those without SCH (413 ± 178 ng/mL vs. 393 ± 255 ng/mL, p=0.036). However, no significant differences in MoM values or NTD risk were detected between the groups. Correlation analysis revealed a low yet significant correlation between AFP levels and hematoma size (r=0.231, p=0.020). ROC analysis revealed that while SCH is a significant factor for elevated MSAFP levels, it exhibits low sensitivity and specificity (AUC: 0.571, 95% CI: 0.507-0.636, p=0.033).

Discussion: SCH is linked to elevated MSAFP levels in patients experiencing threatened abortion in the first trimester. However, SCH does not have a significant impact on MoM values or NTD risk in second trimester screening tests. Clinicians should take SCH into account when interpreting elevated MSAFP levels and monitor patients with larger hematomas more closely. Further research is required to better understanding the clinical implications and improve the management of pregnancies complicated by SCH.

Keywords

Subchorionic Hematoma, Maternal Serum Alpha-Fetoprotein, Threatened Abortion, Second Trimester Screening, Neural Tube Defects

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Corresponding Author: Abdurrahman Alp Tokalioglu, Department of Obstetrics and Gynecology, Faculty of Medicine, University of Health Sciences, Etlik Zubeyde Hanim Women's Health Training and Research Hospital, Ankara, Turkey.
E-mail: alptokalioglu@gmail.com P: +90 532 789 70 96
Corresponding Author ORCID ID: https://orcid.org/0000-0002-1776-2744
Other Authors ORCID ID: Tugrul Basar, https://orcid.org/0000-0003-2028-0647 · Rıza Dur, https://orcid.org/0000-0002-9225-9030
Tufan Arslanca, https://orcid.org/0000-0001-9686-1603 · Mehmet Metin Altay, https://orcid.org/0000-0001-6772-858X
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Introduction

Threatened abortion also known as imminent miscarriage, is characterized by vaginal bleeding and abdominal pain during the first trimester of pregnancy, with a closed cervix and the fetus still viable inside the uterus. It affects approximately 20-25% of all pregnancies. Approximately 20-25% of these patients are found to have a subchorionic hematoma (SCH) during ultrasonographic evaluation. SCH is believed to result from the partial detachment of the chorionic membranes from the uterine wall, leading to an accumulation of blood between the uterine wall and the chorion [1, 2].

The second-trimester screening test typically includes measurements of maternal serum human chorionic gonadotropin (MShCG), maternal serum alpha-fetoprotein (MSAFP), and maternal serum unconjugated estriol (MSuE3). Elevated MSAFP levels are a significant marker for neural tube defects (NTDs), which are severe congenital anomalies of the central nervous system [3]. In addition to NTDs, elevated MSAFP levels may also signal other conditions, such as fetal, placental, and maternal complications, including fetomaternal hemorrhage, which is the most common cause of elevated MSAFP levels aside from NTDs [4].

Alpha-fetoprotein (AFP) is the primary protein produced by the fetal liver and yolk sac and is present in the fetal circulation. It enters the maternal circulation via transplacental diffusion and the amniotic fluid. In non-pregnant women, serum AFP levels are around 1-2 ng/mL, however during pregnancy, MSAFP levels increase by approximately 15% per week from the 12th to the 32nd week, reaching about 500-550 ng/mL at 32 weeks, and then gradually decrease until term [4, 5].

Previous studies have demonstrated an association between subchorionic hematoma in cases of threatened abortion and increased MSAFP levels. Seppala and Ruoslahti reported that 83% of pregnancies with threatened abortion at 13 weeks had elevated MSAFP levels, attributing this increase to fetomaternal hemorrhage [5]. Similarly, elevated MSAFP levels have been observed following invasive procedures like chorionic villus sampling (CVS) and amniocentesis due to secondary fetomaternal hemorrhage [6-9].

Given the potential impact of SCH on MSAFP levels and the associated risks, it is essential to understand the implications for second-trimester screening tests. This study aims to investigate whether the presence of subchorionic hematoma in patients diagnosed with threatened abortion during the first trimester affects MSAFP levels and the outcomes of the second-trimester triple screening tests. The study specifically focuses on the risk assessment for NTDs.

Material and Methods

Study Design and Patient Selection

This retrospective study was conducted at the Early Pregnancy Clinic of Etlik Zubeyde Hanım Women's Health Education and Research Hospital. The medical records of 922 patients who presented with vaginal bleeding and were diagnosed with threatened abortion between August 2013 and August 2015 were reviewed. Inclusion criteria for the study were patients with a single viable pregnancy between 6 and 18 weeks, with no known systemic diseases or bleeding disorders.

A total of 487 patients were excluded: 80 experienced miscarriage, 315 did not return for follow-up, and 92 did not meet the inclusion criteria. Ultimately, 435 patients were eligible for the study, of 102 having subchorionic hematoma (Group 1) and 248 without(Group 2).

Data were collected on patient demographics, including age, body mass index (BMI), gestational age at diagnosis, gravidity, parity, and ultrasound findings. Ultrasonographic evaluations were conducted using a GE Logiq P5 ultrasound machine equipped with both transabdominal and transvaginal probes performed by two obstetricians. The size and location of subchorionic hematomas were recorded. The triple screening test results, including AFP levels, corrected MoM values, and NTD risk, were retrieved from the hospital's information system.

Statistical Analysis

Data analysis was performed using SPSS for Windows version 11.5. Descriptive statistics were calculated for both demographic and clinical characteristics. The Student's t-test was applied to compare normally distributed continuous variables between groups, while categorical variables were analyzed using chi-square test (Pearson, Yates correction, and Fisher's exact test). Correlation analysis was conducted to assess relationships between continuous variables. A p-value <0.05 was considered statistically significant. ROC analysis was performed to determine cut-off values.

Ethics Approval

This study received approval from the Ethics Committee of Etlik Zubeyde Hanım Women's Health Education and Research Hospital (Date: 2016-02-25, No: 205).

Results

Table 1 displays the demographic characteristics of the 350 patients included in the study. The average age of the patients was 27.5 ± 5.6 years, with a mean gestational age of was 10.8 ± 3.1 weeks at the time of diagnosis. Comparing the demographic characteristics between the SCH-positive and SCH-negative groups, the mean age was similar (27.5 ± 5.5 years vs. 27.5 ± 5.7 years, p=0.748). However, BMI was significantly higher in the SCH-negative group than in the SCH-positive group (25.4 ± 4.5 vs. 24.2 ± 3.9, p=0.011).

The average gravidity and parity were similar between the two groups (gravidity: 2.2 ± 1.2 vs. 2.3 ± 1.4, p=0.787; parity: 0.78 ± 0.86 vs. 0.76 ± 0.91, p=0.647). The mean gestational age at diagnosis did not differ significantly between the groups (10.7 ± 3.0 weeks vs. 10.8 ± 3.1 weeks, p=0.920) (Table 1). No significant differences were in age, gravidity, and parity between the groups.

Previous Pregnancy Outcomes and Smoking Status

Comparing previous pregnancy outcomes and smoking status no significant differences were observed between the groups in terms of abortion, ectopic pregnancy, voluntary curettage, or smoking habits.

Triple Screening Test Results

Table 2 presents the results of the triple screening tests. A significant difference in AFP levels was observed between the SCH-positive and SCH-negative groups (413 ± 178 ng/mL vs. 393 ± 255 ng/mL, p=0.036). The mean MoM values were

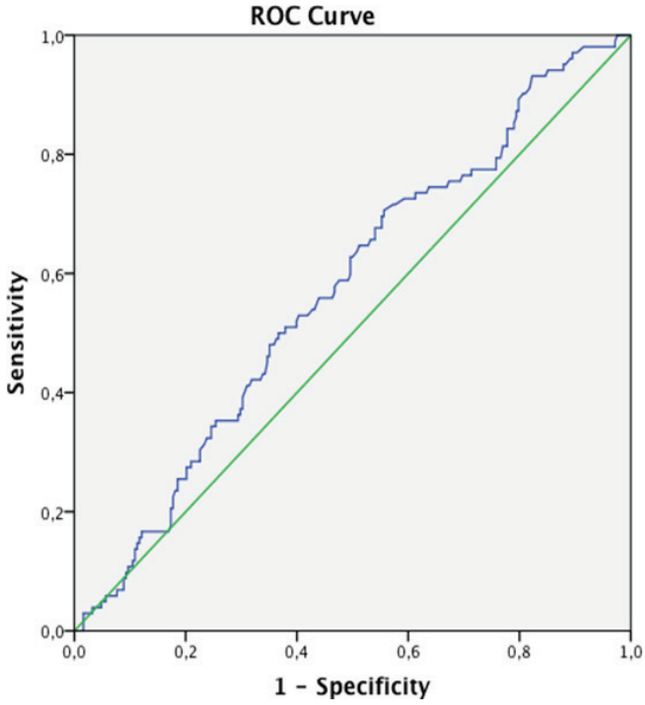

Figure 1. ROC curve of subchorionic hematoma as a variable for elevated MSAFP levels

Table 1. Demographic Characteristics of the Study Population and Comparison Between SCH-Positive and SCH-Negative Groups

Variable	SCH (+) (n=102)	SCH (-) (n=248)	Total (n=350)	p value
Age (years)	27.5 ± 5.5	27.5 ± 5.7	27.5 ± 5.6	0.748
BMI	24.2 ± 3.9	25.4 ± 4.5	25.0 ± 4.4	0.011
Gravida	2.2 ± 1.2	2.3 ± 1.4	2.3 ± 1.3	0.787
Parity	0.78 ± 0.86	0.76 ± 0.91	0.76 ± 0.90	0.647
Gestational Age (weeks)	10.7 ± 3.0	10.8 ± 3.1	10.8 ± 3.1	0.920

BMI: Body Mass Index (kg/m²)
SCH: subchorionic hematoma

Table 2. Triple Screening Test Results

Variable	SCH (+) (n=102)	SCH (-) (n=248)	p value
AFP	413 ± 178	393 ± 255	0.036
MoM	1.1 ± 0.4	1.0 ± 0.7	0.689
Screening Week (weeks)	17.1 ± 0.7	16.9 ± 0.8	0.071
NTD Risk (%)	3 (2.9)	11 (4.4)	0.765
Smoking (%)	5 (4.9)	18 (7.3)	0.568

SCH: subchorionic hematoma
AFP: Alpha fetoprotein (ng/mL)
MoM: Multiple of Median
NTD: Neural Tube Defect

Table 3. Comparison of NTD Risk with MSAFP Cut-off Levels in Study Participants

MSAFP cut-off	Normal NTD Risk	Increased NTD Risk	Total	p value
<2.0 MoM	150 (96.2%)	6 (3.8%)	156	<0.001
≥2.0 MoM	10 (62.5%)	6 (37.5%)	16	<0.001

MoM: Multiple of Median
AFP: Alpha fetoprotein (ng/mL)
NTD: Neural Tube Defect

comparable between the two groups (1.1 ± 0.4 vs. 1.0 ± 0.7, p=0.689). Similarly, the gestational age at the time of screening did not significantly differ between the groups (17.1 ± 0.7 weeks vs. 16.9 ± 0.8 weeks, p=0.071). The NTD risk was similarly comparable between the groups (3% vs. 4.4%, p=0.765).

Correlation analysis revealed a low yet significant correlation between AFP levels and hematoma size (r=0.231, p=0.020). No significant correlation was found between AFP levels and hematoma location (r=0.450, p=0.081). Additionally, no significant correlations were observed between AFP levels and maternal age, gravida, parity, or gestational age.

The comparison of NTD risk with MSAFP cut-off levels is summarized in Table 3. Among patients with MSAFP levels <2.0 MoM, 96.2% had a normal NTD risk, while 3.8% had an increased NTD risk. In contrast, among patients with MSAFP levels ≥2.0 MoM, 62.5% had a normal NTD risk, while 37.5% had an increased NTD risk. This difference was statistically significant (p<0.001).

ROC Curve Analysis

The ROC curve analysis was utilized to evaluate the diagnostic performance of subchorionic hematoma as a variable for elevated MSAFP levels. The area under the curve (AUC) was 0.571 (95% CI: 0.507-0.636, p=0.033), indicating that subchorionic hematoma is a significant predictor of elevated MSAFP levels. However, the variable demonstrates low diagnostic accuracy, with a sensitivity 62.7%, and specificity 51.4% ((Figure 1).

Discussion

This study investigated the impact of subchorionic hematoma (SCH) on MSAFP levels and triple screening test outcomes in patients diagnosed with threatened abortion during the first trimester. Our findings indicate that the presence of SCH is associated with elevated MSAFP levels. However, there were no significant differences in MoM values or NTD risk between patients with and without SCH.

The observed rise in MSAFP levels in patients with SCH aligns with previous studies that have associated elevated MSAFP levels with fetomaternal hemorrhage [10, 11]. For instance, Seppala and Ruoslahti found elevated MSAFP levels in 83% of pregnancies with threatened abortion at 13 weeks, attributing this to fetomaternal hemorrhage [4]. Similarly, Christmas et al. examined the effect of fetomaternal hemorrhage on adverse pregnancy outcomes in patients with elevated second-trimester maternal serum AFP levels and found a significant correlation [11]. In this regard, Lachman et al. reported that the detection and measurement of fetomaternal hemorrhage using serum alpha-fetoprotein and the Kleihauer technique confirmed these observations [7].

Notwithstanding, elevated MSAFP levels have been observed following invasive procedures like chorionic villus sampling (CVS) and amniocentesis due to secondary fetomaternal hemorrhage. Katiyar and colleagues detected fetomaternal hemorrhage following CVS, which was associated with a rise in maternal serum alpha-fetoprotein levels [6]. Moreover, Fuhrmann et al. reported that maternal serum AFP levels increased following CVS due to fetomaternal hemorrhage [8]. Conversely, Makrydimas et al. investigated fetomaternal hemorrhage following coelocentesis and found no significant

impact on AFP levels [13].

Despite the elevated MSAFP levels in patients with SCH, our study did not find significant differences in MoM values or NTD risk between the groups. This suggests that while SCH can elevate MSAFP levels, it may not substantially impact the accuracy of NTD risk assessment using triple screening tests. This finding is crucial for clinical practice as it indicates that SCH should be considered when interpreting elevated MSAFP levels, but it does not necessarily indicate a higher risk of NTDs. This aligns with the work of Maso et al., who observed that the presence of SCH did not significantly alter pregnancy outcomes [9].

The absence of a significant difference in MoM values and NTD risk between SCH-positive and SCH-negative groups could be attributed to several factors. First, the size of the hematoma may play a role. In our study, correlation analysis showed a low but significant correlation between AFP levels and hematoma size ($r=0.231$, $p=0.020$), indicating that larger hematomas are associated with higher AFP levels. However, the overall impact of hematoma size on MSAFP levels and subsequent NTD risk assessment may be limited. This finding is consistent with previous studies that have shown variable impacts of hematoma size on pregnancy outcomes. Similarly, Tuuli et al. conducted a meta-analysis and found that while SCH was associated with adverse pregnancy outcomes, the size of the hematoma was a critical factor in determining the level of risk [14].

Second, the timing of the hematoma's formation and its resolution could influence MSAFP levels. Hematomas that resolve earlier in pregnancy may have less impact on second trimester screening results compared to those that persist. Regan et al. noted that the timing and resolution of hematomas significantly affected pregnancy outcomes, suggesting that early resolution might mitigate some risks [15]. Early resolution of SCH can lead to normalization of MSAFP levels, thereby reducing the potential for false-positive NTD screening results.

Another important consideration is the clinical management of patients with SCH. In our study, the lack of significant differences in adverse pregnancy outcomes such as abortion history, ectopic pregnancy, voluntary curettage, and smoking status between SCH-positive and SCH-negative groups suggests that the presence of SCH alone may not be a decisive factor in determining pregnancy outcomes. This aligns with previous studies that have shown variable impacts of SCH on pregnancy outcomes, with some studies reporting increased risks of adverse outcomes and others finding no significant associations. Stephenson et al., reported similar findings in a study involving 197 couples, where the presence of SCH did not consistently predict adverse outcomes [16].

Additionally, SCH may have a transient effect on MSAFP levels. Wilcox et al. demonstrated that early pregnancy losses are common and often associated with transient elevations in serum markers, including MSAFP [17]. This transient nature might explain why some studies, including ours, do not find long-term impacts of SCH on pregnancy outcomes despite initial elevations in MSAFP levels.

The findings of this study carry several clinical implications. First, the presence of SCH should be taken into account when evaluating elevated MSAFP levels during the second trimester. While elevated MSAFP levels may indicate an increased risk of NTDs, SCH can also contribute to these elevated levels. Therefore, clinicians should take into account the presence of SCH and potentially use additional diagnostic tools such as detailed ultrasound evaluations to accurately assess NTD risk. Alberman emphasized the importance of considering other diagnostic tools in conjunction with MSAFP levels to provide a comprehensive assessment [18]. Second, the correlation between hematoma size and AFP levels suggests that patients with larger SCH should be monitored more closely. These patients may require more frequent follow-up visits and additional diagnostic tests to ensure the well-being of the fetus and to manage any potential complications. Kurki and Ylikorkala suggested that close monitoring and follow-up are crucial in managing pregnancies complicated by SCH [19].

Furthermore, the results of this study underscore the importance of considering other potential factors that could influence MSAFP levels. For example, maternal weight, diabetes, and smoking status are known to affect MSAFP levels and should be taken into account when interpreting screening results. Blumenfeld and Brenner noted that various maternal factors, including thrombophilia and lifestyle choices, significantly influence MSAFP levels [20]. Additional research is necessary to investigate the influence of these factors on MSAFP levels in patients with SCH and to create more comprehensive risk assessment models that include these variables.

This study has several limitations. The retrospective design may introduce selection bias, and the sample size, particularly for patients with SCH, was relatively small. Additionally, the study did not consider other potential factors that might affect MSAFP levels, such as maternal weight, diabetes, and smoking status. Future research should focus on addressing these limitations by conducting larger prospective studies that account for these confounding factors. Additionally, investigating the longitudinal effects of SCH on pregnancy outcomes and MSAFP levels throughout pregnancy could provide valuable insights into the temporal dynamics of these associations. Harlap and Shiono emphasized the importance of prospective studies to gain a deeper understanding of long-term impacts of SCH on pregnancy outcomes [21]. Furthermore, the clinical significance of the low correlation between AFP levels and hematoma size warrants further investigation. Understanding the mechanisms underlying this correlation could provide valuable insights into the pathophysiology of SCH and its impact on pregnancy outcomes. For instance, examining the role of placental vascularization and the degree of fetomaternal hemorrhage in relation to hematoma size could help elucidate the biological processes driving the observed associations. Blumenfeld and Brenner noted the importance of understanding the pathophysiological mechanisms to improve clinical management and outcomes [20].

Conclusion

In conclusion, this study demonstrates that subchorionic hematoma is associated with elevated MSAFP levels in patients with threatened abortion during the first trimester. However, the presence of SCH does not significantly impact MoM values or NTD risk in second trimester screening tests. Clinicians should consider SCH when interpreting elevated MSAFP levels

and monitor patients with larger hematomas more closely. Further research is needed to explore the clinical implications of these findings and to improve the management of pregnancies complicated by SCH.

Scientific Responsibility Statement
The authors declare that they are responsible for the article's scientific content including study design, data collection, analysis and interpretation, writing, some of the main line, or all of the preparation and scientific review of the contents and approval of the final version of the article.

Animal and Human Rights Statement
All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or compareable ethical standards.

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Conflict of Interest
The authors declare that there is no conflict of interest.

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